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Bioavailability of soluble microbial products as the autochthonous precursors of disinfection by-products in aerobic and anoxic surface water

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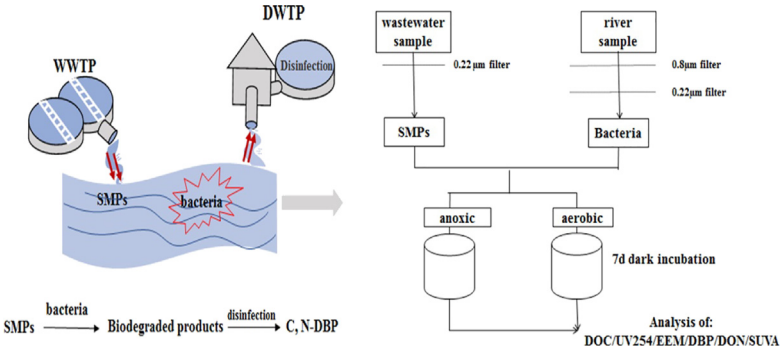
HIGHLIGHTS

- SMPs in the river were biodegraded under aerobic and anoxic conditions.
- Biodegradation of DBP precursors increased with the decrease of dissolved oxygen.
- In the early phase of incubation, DCAN increased as intermediates were formed.
- Anoxic condition promoted the removal of aromatics resulting in a lower DBP yield.
- Dissolved oxygen and SMPs were key factors in shaping microbial compositions.

GRAPHICAL ABSTRACT



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ABSTRACT

Soluble microbial products (SMPs), as a major part of the effluent organic matter discharged into surface water, may affect the formation of disinfection by-products (DBP) in downstream drinking water treatment plants. In this study, excitation emission matrix fluorescence with parallel factor analysis (EEM-PARAFAC), infrared spectroscopy (IR), high performance size-exclusion chromatography (HPSEC) and 16SrRNA high-throughput sequencing were used to investigate the aerobic and anoxic bioavailability of SMPs in surface water and evaluate their influences on DBP formation upon chlorination in a subsequent drinking water plant. In this study, SMPs were utilized by enriched microbial communities such as Bacteroidetes and Proteobacteria, but the accumulation of SUVA was pronounced during the two oxygen conditions. Biodegraded SMPs had higher humic substructures and lower protein-like components. Due to the presence of SMPs, microbial community compositions were influenced during biodegradation. Moreover, DO was the main factor in biodegradation of SMPs, thus affecting a series of processes, such as microbial compositions, properties of SMPs, DBP formation and reactivity. DBP formation potential decreased after anoxic and aerobic incubations. However, SMPs after aerobic degradation had higher DBP reactivity meanwhile the opposite was found for anoxic incubation. Based on the analysis of IR and HPSEC, it was found that some new substrates or intermediates with MW (220 KDa, <1 KDa) during microbial incubation may contribute to the formation of trihalomethane (THMs), chloral hydrate (CH), dichloroacetonitrile (DCAN) and trichloronitromethane (TCNM) in each DBP sampling episode.

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1. Introduction

The Pearl River flows through one of the most developed and densely populated areas in China, and it acts as both a wastewater disposal sink and potable water source. As a result, effluent organic matter (EfOM) discharged into the river from sewage treatment plants is transported some distance and then enters downstream drinking water treatment plants (DWTPs) (Chen et al., 2009). EfOM is rich in soluble microbial products (SMPs) (Xie et al., 2016) which may react with disinfectants to produce genotoxic and cytotoxic carbonaceous (C-DBPs) and nitrogenous (N-DBPs) disinfection by-products (Krasner et al., 2009). N-DBPs deserve particular attention because their nitrogen-based structure is far more toxic than that of C-DBPs, and therefore threatening to human health (Krasner et al., 2013). SMPs are organic macromolecules released by microorganisms during biological sewage treatment. Xie and his colleagues found that SMPs account for 61% of the chemical oxygen demand in the wastewater treatment plant they studied (Xie et al., 2016). They are classified into two groups based on the bacterial phase: 1) utilization-associated products (UAP) containing small carbonaceous compounds associated with biomass growth; 2) biomass-associated products (BAP), containing cellular macromolecules derived from biomass decay (Barker and Stuckey, 1999). Previous studies have reported that small-molecule SMPs as the majority of DBP precursors may increase disinfection by-products in the subsequent disinfection process of DWTPs (Chen et al., 2009; Golea et al., 2017). Therefore it is important to understand the fate of SMPs before they enter DWTPs.

To minimize undesired effects, biodegradation has been suggested as a way to remove SMPs from the effluent of wastewater treatment systems (Dong et al., 2013). A group led by Kim has shown (Kim et al., 2016) that an enriched microbial community could effectively degrade SMPs if the operating conditions are carefully controlled, and key microbial populations involved in SMPs degradation were identified in an anaerobic reactor. In general, most studies were performed with synthetic water (not with river water) or with bacteria inoculated from wastewater reactors (not with microorganisms from the river).

In addition, the natural aerobic biotransformation of dissolved organic matter (DOM) as a heterogeneous precursor of DBP in surface water has been widely studied (Chow et al., 2009; Mostofa et al., 2011), but the linkages between natural transformation processes and autochthonous precursors of DBP have received relatively little scholarly attention. Reports from Bastviken's group have shown that anoxic conditions are common in many aquatic ecosystems, especially in poorly mixed water and surface sediments. Hence, aquatic bacteria are likely to be exposed to both aerobic and anoxic conditions in rivers, causing inconsistent bioavailability of organic matter (Bastviken et al., 2001; Bastviken et al., 2003). Therefore, the role of aerobic and anoxic degradation may be important in the fate of DBP precursors. However, up until now, few studies about their processes have been reported. It remains unclear how different redox conditions alter autochthonous organics concentrations and properties, affecting the formation of DBPs.

In this study, laboratory experiments were designed to investigate the aerobic and anoxic bioavailability of SMPs using river water as the inoculum. The objectives of this study were: (1) to compare the effect of bacteria from the surface water on SMPs under aerobic and anoxic conditions; (2) to evaluate the changes in DBPs after biodegradation; (3) to understand the relationship between the bacterial composition and SMPs degradation.

2. Materials and methods

2.1. SMPs collection

A lab-scale sequencing batch reactor (SBR) was simulated to produce the SMPs tested. Active sludge was collected from a secondary sedimentation tank of the Lijiao wastewater treatment plant in Guangzhou, China. The sludge was cleaned for 12 h with tap water to eliminate heterogeneous dissolved organic matter. Glucose (800 mg/L) was added to the solution as the only organic substrate; other components (in mg/L) of the synthetic wastewater were NH_4Cl, 69; KH_2PO_4, 35; $NaHCO_3$, 0.24; $CaCl_2.5H_2O$, 0.37; $MgSO_4.7H_2O$, 5.07; $MnCl_2 \cdot 4H_2O$, 0.275; $ZnSO_4 \cdot 7H_2O$, 0.44; $CoCl_2 \cdot 6H_2O$, 0.42 and $Na_2MoO_4 \cdot 2H_2O$, 1.26. The dynamics of SMPs production were reported by Wu et al. (2018). Utilization-associated products (UAP) were first produced during substrate utilization and biomass growth phase and then further biodegraded after 3.5 h of incubation. During this organic depletion and biomass decay phase, biomass-associated products (BAP) began to increase after hour 46. After aerobic incubating for 130 h at 25 °C, the nutrient substrates were completely exhausted by the microorganisms, leaving only autochthonous SMPs in the solution. The SMPs solution was obtained through a 0.22 μm mixed cellulose filter to remove over 99.5% of the bacteria and suspended solids. It was stored in the dark at 4 °C until analysis within a week.

2.2. Sampling site and surface water collection

The surface waters were sampled from the Pearl River in the south of China, which usually receives biologically treated municipal wastewater discharged from sewage treatment plants. With large inputs of wastewater and organic pollutants, anoxic conditions were common in many tributaries of the Pearl River, especially in polluted rivers. The concentration of dissolved oxygen was observed from 0.8 to 8.2 mg/L along the Pearl River estuary and the near-shore open water site (Li et al., 2018). The sampling location was established below the Lijiao wastewater treatment plant: The tributary of the Pearl River (113°22′443″E, 23°3′600″N) is located in the south of Guangzhou, China and it receives large wastewater volumes containing SMPs before entering downstream drinking water treatment plants. Samples were collected using a water-harvesting sampler. The temperature, pH and dissolved oxygen of surface water (as shown in Table S1) were measured in situ using a thermometer, pH meter and dissolved oxygen meter (HACH, USA). Samples were collected in 25 L containers and immediately transported to lab. Within 2 days, surface waters were filtered through 0.8 μm mixed cellulose filter to remove large particles of material and animals, and then enriched 20-fold on 0.22 μm mixed cellulose filter by a vacuum filter. Eventually 0.22 μm filter with bacteria was suspended in filtered river water, which was defined as the inoculum solution (Hur et al., 2011). This procedure yielded approximate population densities of bacteria, as measured in situ by Luria-Bertani medium plate counting (as shown in Table S1).

2.3. Experimental setup

Biotransformation experiments were conducted based on OECD Test Guideline No. 301 with some modifications (OECD, 2004). For microbial experiments, nine flasks were divided into three treatments, each in triplicate. The treatments were the aerobic incubation, anoxic incubation and control experiment and all flasks were shaken at 150 rpm per minute in the dark at 25 °C for 15 days. The aerobic and anoxic experiments were conducted using 2.5 L sterile conical flasks, each including 150 mL of the inoculum solution and 1500 mL of the SMPs solution. For aerobic incubation, each flask was sealed with breathable sealing film to allow oxygen infiltration. For anoxic incubation, the headspace of each flask was repeatedly vacuumed and filled with nitrogen for 1 to 2 h daily before being sealed with a para film. The concentrations of dissolved oxygen (DO) in the anoxic containers were measured to maintain below 2 mg/L with portable dissolved oxygen meter (HACH, USA) (as shown in Fig. S1). To check the role of other mechanisms (sorption, hydrolysis), evaluate the activity of the bacteria and measure the carbon evolution of the inoculum, the corresponding control experiments were as follow. i) Abiotic control: 1% sodium azide was added to a mix solution (SMPs and inoculum solution at a ratio of 10:1) in order